ART 34 AMDT

SUB A1

CLAIMS

1. A method of obtaining a sample of a biomolecule from a suspension comprising a solution containing the biomolecule and insoluble material, the method comprising the steps of:

(a) providing a biomolecule purification assembly comprised of a vessel having a liquid inlet and of a filter unit removably located on the liquid inlet;

(b) effecting a filtration of the suspension through the filter unit so as to cause the solution to enter the vessel through the liquid inlet;

(c) removing the filter unit from the liquid inlet;

(d) immobilising the biomolecule on a solid phase support; and

(e) subjecting the biomolecule to at least one of the steps of washing on the support and elution from the support to obtain a purified sample of the biomolecule.

2. A method as claimed in claim 1 wherein the biomolecule is a nucleic acid.

3. A method as claimed in claim 2 wherein the nucleic acid is DNA.

4. A method as claimed in claim 3 wherein the DNA is plasmid DNA.

5. A method as claimed in any one of claims 1 to 4 wherein the step (e) comprises washing the solid phase support and said support is contained within the vessel during this step.

AMENDED SHEET

6. A method as claimed in claim 5 wherein step (e) comprises eluting the biomolecule from solid phase support contained within the vessel, the elution being effected through said inlet of the vessel.

Sub A1

7. A method as claimed in any one of claims 1 to 6 wherein the step (b) the suspension to be filtered is contained in a well and in step (c) the filter unit is discharged into that well.

8. A method as claimed in any one of claim 1 to 7 wherein the biomolecule is absorbed onto the solid phase support in the presence of a binding agent for effecting said adsorption.

9. A method as claimed in claim 8 wherein, for the purposes of step (d) the filtrate in the vessel contains the binding agent and the filtrate is discharged onto the support and the resultant mixture is taken back into the vessel.

10. A method as claimed in claim 8 wherein, for the purposes of step (d), the filtrate is discharged into a mixture of the support and the binding agent and the resultant mixture is taken back into the vessel.

11. A method as claimed in any one of claims 1 to 10 wherein the solid phase support comprises magnetic beads.

12. A method as claimed in any one of claims 1 to 11 wherein the vessel of the biomolecule purification assembly is an open-ended, vertically disposed column.

13. A method as claimed in claim 12 wherein the column has an upper bore section and a lower bore section of reduced diameter as compared to the upper section.

14. Apparatus for obtaining a sample of a biomolecule from a suspension comprising a solution of the biomolecule and insoluble material the apparatus comprising

(i) a filtration station at which is provided a biomolecule purification assembly comprised of a vessel having a liquid inlet and a removable filter unit located at the inlet of the vessel, said filtration station being provided with means for causing the solution present in the suspension to pass through the filter unit and the liquid inlet into the vessel;

(ii) means for removing the filter unit from the vessel; and

(iii) at least one of a washing station and an elution station.

15. Apparatus as claimed in claim 14 comprising an elution station and a washing station.

16. Apparatus as claimed in claim 14 or 15 further comprising a solid phase support supply station preferably provided between (ii) and (iii).

17. An apparatus as claimed in any one of claims 14 to 16 wherein the vessel is an open-ended flow-through column.

18. An apparatus as claimed in claim 17 wherein the column has an upper bore section and a lower bore section of reduced diameter compared to the upper section.

19. An apparatus as claimed in claim 17 or 18 wherein the filter unit comprises a sleeve which incorporates the filter and which is located over the lower end of the column.

20. An apparatus as claimed in claim 19 wherein the upper surface of the filter has a depression and the lower end of the column locates in the depression.

21. An apparatus as claimed in any one of claims 14 to 20 in which the pore size of the filter is in the range 0.2 to 50 microns.

22. An apparatus according to any one of claims 14 to 21 in which the suspension to be filtered is contained in a well and once the filtration operation is complete the filter unit is automatically discarded into that well.

23. An apparatus according to claim 22 further comprising a stripping arrangement which acts on the filter unit when the assembly is moved upwards causing the filter unit to be discharged into the well.

24. An apparatus according to any one of claims 14 to 23 which is capable of handling an array of biomolecule purification assemblies.

25. An apparatus according to claim 24 wherein each individual step of filtration, removal of the filtration unit washing and elution is adapted to be effected simultaneously on all members of the array.

26. An apparatus as claimed in claim 14 comprising

(a) a head arrangement having a plurality of individual column supporting heads on which the upper ends of disposable columns may be removably mounted and which have fluid flow passageways for transfer of fluids into and out of the upper ends of the columns;

(b) means for moving a supply of disposable columns so that upper ends of columns to be mounted on the supporting heads are presented below said heads;

AMENDED SHEET

(c) means for moving the head arrangement relatively downwardly towards the tops of the columns whereby the upper ends thereof become removably mounted on said head, and for moving the head arrangement relatively upwardly with columns mounted thereon in readiness for subsequent stages of the process.

(d) reservoirs for reagents/wash solutions as appropriate;

(e) means (e.g. for providing pressure variation in the columns) for causing liquid samples, liquid extracts or mixtures of such liquids with reagents thereof, provided at the lower tips of the columns (e.g. in wells of a microlite plate, BioBlock or similar) to be drawn upwardly into the columns and discharged from the lower tips thereof for processing steps as required to obtain the desired biomolecule; and

(f) means for removing the columns from the heads for disposal.

27. A biomolecule purification assembly comprising a vessel having a liquid inlet and a filter unit removably located on the liquid inlet wherein the vessel is in the form of a column having a first bore section and a second bore section of reduced diameter as compared to the first section, and the filter unit is comprised of a sleeve which houses the filter and which is removably located on the end of the vessel remote from the first bore section.

28. A biomolecule purification assembly as claimed in claim 27 wherein the surface of the filter adjacent to the inlet of the vessel has a depression and the end of the vessel locates in the depression.

29. A filter unit for a biomolecule purification assembly, said filter unit comprising

(a) an elongate sleeve having one end (the "suspension inlet end") through which a suspension to be filtered enters the sleeve and an opposite end

Sub A1

for location over a column of a biomolecule purification assembly, said sleeve having a first body portion and a second body portion which internally tapers from the first body portion to said suspension inlet end, and

(b) a filter having a head portion locating in, and occupying the cross-section of, the first body portion, and a frustoconical or conical body portion extending into said second body portion of the sleeve and tapering therein at a larger angle than the internal taper of the sleeve.

30. A biomolecule purification assembly comprising

(i) an elongate vessel in the form of a column having first and second open ends with a filtrate inlet being provided at the first end of the column, and

(ii) the filter unit as defined in claim 29 removably located over the first end of the vessel.

31. An assembly as claimed in claim 30 wherein the filter unit has an upper depression and said filtrate inlet of the elongate vessel is located in the region of said depression.

32. An assembly as claimed in claim 30 or 31 wherein said elongate vessel is as defined in claim 24.

33. A method as claimed in claim 1 wherein the suspension is obtained by the steps of:

(i) providing a re-suspension of a bacterial pellet in solution;

AMENDED SHEET

(ii) adding lysis solution to the re-suspension to effect release of cellular contents; and

(iii) adding neutralising solution to the lysed re-suspension to effect the formation of a suspension comprising a solution of a biomolecule and insoluble material for use in the method of claim 1;

in which the lysis solution and/or neutralising solution are each added in multiple aliquots which are ejected downwardly into the re-suspension to provide the suspension in the absence of additional mechanical agitation or stirring without the dispensing apparatus coming into contact with the sample or the containing vessel.

34. A method of operating an apparatus as defined in claim 26 using the steps of lysing the cell sample and obtaining the biomolecule from the lysate wherein reagents for lysis are ejected downwardly through the supporting heads into the cell sample to be lysed (either prior or subsequent to mounting columns on the heads).

35. A method as claimed in claim 34 wherein the lysis reagents comprise a lysis solution and a separate neutralising solution.

36. A method as claimed in any one of claims 33 to 35 wherein the cell samples to be lysed are contained within the individual wells of a microtitre well tray or the like and the lysis reagents are ejected into the individual wells.

37. A method as claimed in claim 36 wherein the apparatus comprises a single row of n column supporting heads and the cell samples to be processed are provided in an (m x n) array of microtitre wells wherein the samples in the n wells of any one of the m rows are simultaneously treated with lysis reagents by downward ejection from the n supporting heads.

38. A method as claimed in claim 37 wherein the lysis reagents are discharged into all wells of the (m x n) array prior to disposable columns being mounted on the heads for subsequent steps of the extraction procedure.

39. A method as claimed in any one of claims 33 to 38 wherein the time of dispense for the lysis reagent into a cell sample is 0.1 to 1 second.

40. A method as claimed in any one of claims 33 to 39 wherein the discharge outlet of the column supporting heads is 40mm to 100mm above the surface of the sample into which the lysis reagents are to be discharged.

41. A method as claimed in claim 40 wherein said discharge outlet is about 70mm above said surface of the sample.

42. A method as claimed in any one of claims 33 to 41 wherein the discharge outlet of each supporting head has a diameter of 0.1 to 1mm.

43. A method as claimed in claim 42 wherein said diameter is about 0.75mm.